#### Digeneans (Trematoda) of Freshwater Fishes from Nagano Prefecture, Central Japan

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Abstract Examination of digeneans (Trematoda) parasitizing freshwater fishes collected in Nagano Prefecture, central Japan, revealed that 22 species including two new species occur in this prefecture. Sanguinicola ugui sp. nov. (Sanguinicolidae) is described from the blood vessels of Tribolodon hakonensis (Günther) (Cyprinidae). Azygia rhinogobii sp. nov. (Azygiidae) is described from the stomach of Rhinogobius sp. (Gobiidae, type host) and Gymnogobius urotaenia (Hilgendorf) (Gobiidae), and the intestine of T. hakonensis. Phyllodistomum anguilae Long and Wai, 1958, P. mogurndae Yamaguti, 1934, P. parasiluri Yamaguti, 1934 (Gorgoderidae), and Pseudexorchis major (Hasegawa, 1935) Yamaguti, 1938 (Heterophyidae) are redescribed. The generic diagnosis of the genus Pseudexorchis Yamaguti, 1938 is amended in part. New host and locality records are provided for 20 known species. An outline of the life cycle of Asymphylodora macrostoma Ozaki, 1925 (Lissorchiidae) is given. A furcocystocerous cercaria, probably the cercarial stage of A. rhinogobii sp. nov., is briefly described from Sinotaia quadrata histrica (Gould) (Gastropoda, Viviparidae).

**Key words:** digenean, parasite, new species, furcocystocercous cercaria, taxonomy, life cycle, freshwater fish, Nagano, Japan.

#### Introduction

Since Yamaguti (1934) recorded Allocreadium gotoi (Hasegawa and Ozaki, 1926) Macrolecithus) from Misgurnus anguillicaudatus (Cantor) (Cobitidae) caught in Komi, most presumably in Nagano Prefecture, central Japan, 14 species of adult and one species of immature digeneans have been reported from freshwater fishes in this prefecture (Shimazu, 2003a, b). However, surveyed areas and fishes were limited, and our knowledge of the digenean fauna of freshwater fishes in the prefecture is still incomplete. For example, a lophocercous-brevifurcate-apharyngeate cercaria of the genus Sanguinicola Plehn, 1905 has been known to occur in a viviparid snail in the prefecture (Shimazu, 1979b), but the adult remains to be discovered (Shimazu, 2003b).

In order to obtain further knowledge of the digenean fauna, I surveyed freshwater fishes at various sites in Nagano Prefecture, and have collected considerable number of digeneans. The life cycle of *Asymphylodora macrostoma* Ozaki, 1924 was also studied in the field and laboratory.

This paper describes two new species and adds new records and information on host, locality, and morphology of 20 known species from Nagano Prefecture, with a brief note on the life cycle of *A. macrostoma*.

#### **Materials and Methods**

Freshwater fishes were collected in Nagano Prefecture at irregular intervals and examined fresh for digeneans under a stereoscopic microscope. Main sampling sites of the fishes and the periods were as follows: Hiroi River at Kotobuki, Iiyama, from 1995 to 2004; Torii River at Mure from 1987 to 1995; Lake Kizaki at Oomachi from 1976 to 1994; Nogu River at Oomachi in 1987; Lake Suwa at Suwa from 1976 to 1999; and Tenryu River at Ina in 2000. Gobiids called

"yoshinobori" in Japanese from several sites remain unidentified to species and are referred to as *Rhinogobius* sp. in this paper. It is uncertain whether they all belong to a single species. Prevalence and intensity of infection of each parasite species were not counted.

Digenean specimens were treated as follows: slightly flattened under coverslip pressure, fixed with 70% ethanol, and stained with Grenacher's alum carmine; slightly flattened, fixed with AFA, and stained with Heidenhain's iron hematoxylin; or fixed with hot 10% neutralized formalin and stained with Mayer's hematoxylin. These stained specimens were mounted in Canada balsam. Some were observed alive for study of the excretory system. Some others and some infected organs of hosts were fixed with 10% neutralized formalin, made into serial paraffin sections (10  $\mu$ m thick), and stained with hematoxylin and eosin.

For comparison, institutional specimens were borrowed from the Collections of Dr. Satyu Yamaguti and Dr. Yoshimasa Ozaki deposited in Meguro Parasitological Museum (MPM), Tokyo; National Museum of Nature and Science, Tokyo (NSMT); U.S. National Parasite Collection (USNPC), Beltsville, Maryland, U.S.A.; and the Parasite Collection of the National Museum of Nature (NMNPC), Ottawa, Canada.

Drawings were made with the aid of a camera lucida. Measurements (length by width) are given in millimeters unless otherwise stated. Representatives of the specimens studied have been deposited in the NSMT and USNPC.

The life cycle of *Asymphylodora macrostoma* Ozaki, 1924 was studied in the Torii and Hiroi rivers and in the laboratory.

Class Trematoda
Subclass Digenea
Family Sanguinicolidae
Sanguinicola ugui sp. nov.
(Figs. 1-6)

Type host. Tribolodon hakonensis (Günther)

(Cyprinidae).

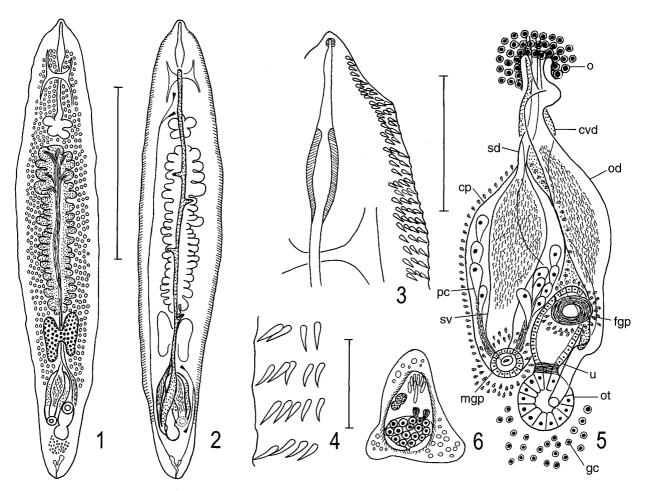
Sites of infection. Blood vessels chiefly of the gills and rarely of the liver, kidneys, and heart. Worms were found at least in the efferent branchial arteries of the gills (NSMT-Pl 5283) and the lumen of the ventricle of the heart (NSMT-Pl 5291). The exact site of infection in the other organs could not be determined.

Localities. Hiroi River (type locality) at Kotobuki (36°54′N, 138°21′E), Iiyama; Lake Suwa at Suwa (36°03′N, 138°06′E); and Tenryu River at Ina (35°50′N, 137°57′E), all in Nagano Prefecture, central Japan.

Specimens deposited. Holotype (NSMT-Pl 5284) and 27 paratypes (NSMT-Pl 5284–5286 and 5288) from Hiroi River on 20 July 1996, 23 October 1996, 24 November 1996, and 5 November 2004; and many vouchers (NSMT-Pl 5283–5295, 5296 and 5297, and 5298) from Hiroi River from 1996 to 2004, Lake Suwa on 2 August 1996 and 5 October 1996, and Tenryu River on 9 September 2000, respectively.

Etymology. The specific name "ugui" is the Japanese common name of the type host.

Description. Based on holotype (NSMT-Pl 5284) and 9 paratypes (NSMT-Pl 5284-5286 and 5288). Body flat, elongate, widest at level of anterior part of testis, pointed at anterior end, gradually narrowing posteriorly and rounded at posterior end, 1.29-1.67 by 0.19-0.35, possibly bearing fine setae (Figs. 1 and 2). Anterior proboscis absent. Tegumental spines lanceolate, weakly curved, arranged in ventrolateral transverse rows from near anterior end of body to middle level of cirrus pouch; 1 each in anterior 1st-4th to 6th rows, thick, 6–8  $\mu$ m long; 2–5 (usually 4) each in remainder, slender,  $14-22 \mu m$  long (Fig. 3). Nerve chords conspicuous; transverse nerve commissure 0.14–0.19 or 10–12% of body length from anterior end. Oral and ventral suckers absent. Mouth small, only slightly subventral, close to anterior tip of body. Small globular sphincteror sucker-like structure about 6–9 μm in diameter encircling esophagus immediately adjacent to mouth aperture (Fig. 3). Esophagus narrow, slender, forming thick-walled fusiform structure (Fig.



Figs. 1–6. Sanguinicola ugui sp. nov. 1, holotype, entire body, dorsal view; 2, holotype, entire body, excretory organs added from free-hand sketches, vitelline follicles omitted, ventral view; 3, paratype, anterior part of body, showing tegumental spines, a sphincter- or sucker-like structure encircling esophagus, and a thick-walled, fusiform structure of esophagus, ventral view; 4, paratype, tegumental spines, ventral view; 5, holotype, terminal genitalia, dorsal view; 6, fully-embryonated egg detected in host's liver. cvd, common vitelline duct; cp, cirrus pouch; fgp, female genital pore; gc, gland cells; mgp, male genital pore; o, ovary; od, oviduct; ot, ootype; pc, prostatic cells; sd, sperm duct; sv, seminal vesicle; u, uterus. Scale bars: 0.5 mm in Figs. 1 and 2; 0.2 mm in Figs. 3 and 5; 0.025 mm in Figs. 4 and 6.

3) 0.06–0.08 by 0.02–0.04 in front of transverse nerve commissure. Intestine X-shaped; ceca very short, usually 4 or rarely 5 or 6 in number, 0.28–0.36 or 21–23% of body length from anterior end. Testis single, median, between ovary and intestinal ceca, 0.44–0.60 by 0.09–0.19, with 21–25 lateral lobes on either side (Figs. 1 and 2). Spermatozoa flowing posteriorly in dorsal median bundle within testis from anterior end of testis to posterior; several similar short bundles from lateral lobes joining this bundle in places; these bundles apparently lacking any kind of duct (Fig. 1). Single diagonal sperm duct running posteriorly from median point of posterior margin of testis

to cirrus pouch, ventral to ovary (Fig. 5). Cirrus pouch fusiform or club-shaped, thin-walled, 0.14–0.16 by 0.05–0.07, directed posteriorly, slightly diagonal, surrounded by small gland cells (Fig. 5). Male genital pore dorsal, sinistrosubmedian, some distance from posterior end of body, lined with cuboidal cells arranged in a single layer, surrounded by small gland cells (Figs. 1, 2, and 5). Seminal vesicle very thin-walled. Small globular pars prostatica may be differentiated immediately before cirrus. Cirrus very short, eversible. Prostatic cells large. Ovary bilobed laterally in shape of butterfly, 0.12–0.17 by 0.09–0.14, median, just behind testis; isthmus

0.85–1.16 or 65–71% of body length from anterior end (Figs. 1, 2, and 5). Oviduct originating from posterior margin of ovarian isthmus, passing posteriorly, making loop, dorsal to sperm duct, including spermatozoa (or acting as oviductal seminal receptacle). Seminal receptacle and Laurer's canal absent. Ootype spherical, lined with columnar cells arranged in single layer, 0.03–0.04 in diameter, almost median, posterior to male genital pore. Large gland cells seen posterior to ootype, possibly not emptying into ootype. Uterus clavate, very short, 0.06–0.10 by 0.03-0.05, directed anteriorly, oblique, bending dorsally at anterior end, lined with cuboidal cells arranged in single layer, surrounded by small gland cells, with well-developed sphincter around aperture; metraterm not seen. Single sphincter present between ootype and uterus. Female genital pore dorsal, dextro-submedian, anterior to male genital pore, surrounded by small gland cells (Figs. 1, 2, and 5). Eggs triangular, not operculate, not embryonated, one in ootype and 1–7 in uterus, if present, 22–34 by  $16 \mu m$ (collapsed in balsam). Vitelline follicles small, profuse from near anterior end of body to cirrus pouch, present laterally to nerve chords, almost confluent anteriorly, separated posteriorly (Fig. 1). Common vitelline duct single, ventral to transverse nerve commissure, intestinal ceca, testis, and ovary; short vitelline ducts joining this duct in places (Fig. 2); vitelline reservoire postovarian, ventral to sperm duct, cirrus pouch, and uterus, uniting with oviduct to form common duct entering ootype dorsally (Fig. 5). Excretory vesicle V-shaped, small, posterior to ootype; right arm longer than left; excretory pore single, posteroterminal (Figs. 1 and 2).

Eggs. Uterine eggs were not yet embryonated. Fully-embryonated eggs were detected in the tissue of the liver, heart, and kidneys (NSMT-Pl 5294) but curiously never in the gills. They were 40–48 by 34–40  $\mu$ m in life, and the miracidia were 30–32 by 18  $\mu$ m in life (Fig. 6).

Excretory system. Arranged asymmetrically (Fig. 2). Right common collecting canal long, but left short; each divided into 2 short collecting

canals each with flame cell; right flame cells between transverse nerve commissure and intestinal ceca, but left near cirrus pouch; flame cell formula 2[(1+1)]=4.

Discussion. Sanguinicola ugui sp. nov. is morphologically characterized by an X-shaped intestine with usually four, very short ceca; the testis with many lateral lobes (21-25 on either side of the body); a butterfly-shaped ovary; an asymmetrically arranged excretory system; and triangular eggs. This new species most closely resembles S. megalobramae Li, 1980 in Megalobrama amblycephala Yih (Cyprinidae) from China (Li, 1980) in body size (1.23–1.74 long in the latter species), number of the lateral testicular lobes (18–22 on either side in the latter species), and the asymmetrically arranged excretory system. However, the new species is different from the latter species in having four intestinal ceca instead of a single cecum, in forming triangular instead of oval eggs, and in lacking long setae at the posterior part of the body.

Sanguinicola idahoensis Schell, 1974 in Oncorhynchus mykiss (Walbaum) (syn. Salmo gairdneri Richardson) (Salmonidae) from U.S.A. also has the asymmetrically arranged excretory system (Schell, 1974) The new species is distinguished from S. idahoensis in having more lateral testicular lobes (21–25 instead of 14–18 on either side), a thin-walled seminal vesicle, and a cirrus pouch; and in forming triangular instead of oval eggs.

The following species also form triangular eggs: *S. armata* Plehn, 1905, *S. chalmersi* Odhner, 1924, *S. clarias* Imam, Marzouk, Hassan and Itman, 1984, *S. huronis* Fischthal, 1949, *S. inermis* Plehn, 1905, *S. intermedia* Ejsmont, 1926, *S. lungensis* Tang and Ling, 1975, *S. rhodei* Wang, 1983, *S. rutili* Simón-Martín, Rojo-Vázquez and Simón-Vicente, 1988, *S. sanliensis* Wang, 1982 (originally spelt *sanliense*), and *S. shantsuensis* Lung and Shen, 1965 (Ejsmont, 1926; Fischthal, 1949; Imam *et al.*, 1984; Lung and Shen, 1965; Odhner, 1924; Plehn, 1905; Simón-Martín *et al.*, 1988; Tang and Ling, 1975; Wang, 1982, 1983). However, these species have less than 20 lateral

testicular lobes on either side of the body. Sanguinicola magnus Hu, Long and Lee, 1965 has a larger body size (1.95–2.93 long) and more lateral testicular lobes (27–29 on either side) and forms oval eggs (Hu et al., 1965). The shape of eggs has not yet been described in S. lophophora Erickson and Wallace, 1959, S. occidentalis Van Cleave and Mueller, 1932, S. skrjabini Akhmerov, 1960, and S. platyrhynchi Guidelli, Isaac and Pavanelli, 2002. The first three have less than 20 lateral testicular lobes on either side (Akhmerov, 1960; Erickson and Wallace, 1959; Van Cleave and Mueller, 1932). The last one has a smooth tegument and six intestinal ceca (Guidelli et al., 2002), but the number of the lateral testicular lobes on either side has not been described in it.

A cercaria of lophocercous-brevifurcate-apharyngeate type (NSMT-Pl 5299), which is considered to be a cercaria of Sanguinicola (see Shimazu, 2003b), was found to develop in a globular (most presumably daughter) sporocyst in Semisulcospira libertina (Gould) and Se. dolorosa (Gould) (Gastropoda, Pleuroceridae) in the Hiroi River in 1999. This cercaria has two-paired flame cells arranged asymmetrically as seen in the above-described adult and a single caudal excretory canal, which is forked at the tail bifurcation to open outside at the tip of each tail furca. Judging from the morphology and host snail species, the cercaria can be identical with Cercaria andoi Faust, 1924, which was redescribed by Ito (1964). It is possible that the cercaria may be that of the present new species. However, several experimental attempts to infect the cercaria to T. hakonensis have not been successful (my unpublished data). In Lake Suwa, on the other hand, an unidentified cercaria of the same type has been known from Sinotaia quadrata histrica (Gould) (Gastropoda, Viviparidae). The excretory system of this cercaria also is the same as that of the above-mentioned adult and cercaria (Shimazu, 1979b).

The flame cell formula is 2[(1+1)]=4 in the above-mentioned two cercariae, the present new species, *S. idahoensis*, and *S. megalobramae* (this paper; Schell, 1974; Li, 1980). That of the cer-

caria of *S. armata* Plehn, 1905 was described as 2[2+1]=6 [or 2[(1+1)+1]=6] by Tang *et al.* (1986, fig. 1) and as 2[(1+1)+(1+1)]=8 by Sendersky and Dobrovolsky (2004). Evidently, these formulae are questionable and need confirmation.

With regard to the gender of the generic name Sanguinicola Plehn, 1905, Yamaguti (1971) and Nolan and Cribb (2005, p. 99, footnote) treated it as masculine, emending the spellings of the adjectival specific names of five known species of the genus from feminine to masculine (for example, from armata to armatus). However, I regard the gender as feminine in accordance with ICZN Art. 30.1.4.2 (Anonymous, 1999) because, when establishing the new genus Sanguinicola, Plehn (1905) originally treated it as feminine by combining the generic name Sanguinicola with the adjectival specific name armata (feminine). The original spellings of the adjectival specific names of the five species are correct and retained, and the emended spellings are incorrect subsequent spellings. The specific name maritimus Nolan and Cribb, 2005 (masculine) should be changed to maritima (feminine) in accordance with ICZN Art. 34.2.

# Family Azygiidae Azygia rhinogobii sp. nov. (Figs. 7–11)

Hosts. Rhinogobius sp. (Japanese name "yoshinobori") (type host), Gymnogobius urotaenia (Hilgendorf) (Gobiidae), and Tribolodon hakonensis (Cyprinidae).

Sites of infection. Stomach of Rhinogobius sp. and G. urotaenia and intestine of T. hakonensis.

Type locality. Lake Suwa at Suwa (36°03′N, 138°06′E), Nagano Prefecture, central Japan.

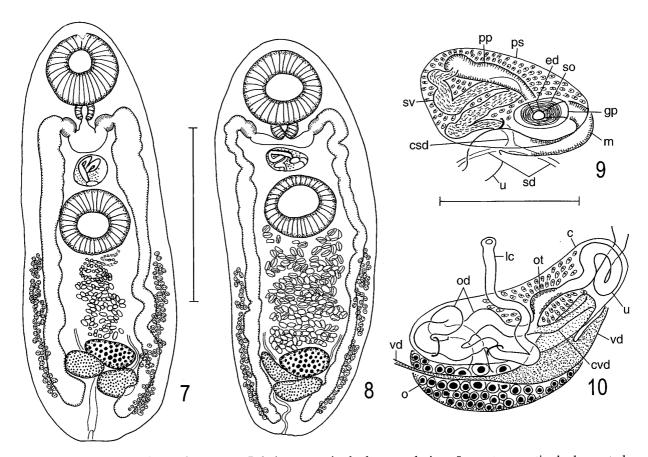
Specimens deposited. Holotype (NSMT-Pl 5300) and 1 paratype (NSMT-Pl 5301) from *Rhinogobius* sp. on 20 November 1993 and 30 October 1993, respectively; 1 paratype (NSMT-Pl 5306) from *T. hakonensis* on 5 October 1991; 9 immature vouchers (NSMT-Pl 5301–5305) from *Rhinogoius* sp. on 24 September 1992, 23 Octo-

ber 1992, 2 October 1993, and 2 August 1996; and 4 immature vouchers (NSMT-Pl 5307–5309) from *G. urotaenia* on 16 October 1993, 30 October 1993, and 20 November 1993.

Etymology. The specific name "rhinogobii" is derived from the generic name "Rhinogobius" of the type host.

Description. Based on 3 adult type specimens. Body elongate, 2.30–2.83 by 0.88; forebody 1.28–1.39 long, occupying 43–51% of body length (Figs. 7 and 8). Tegument smooth. Oral sucker subterminal, 0.40–0.52 by 0.40–0.49. Prepharynx absent. Pharynx elliptical, 0.13–0.16 by 0.11–0.14. Esophagus short, inverted T- or Y-shaped. Intestinal ceca slightly undulating, extending to near posterior end of body. Ventral sucker equatorial or slightly pre-equatorial, 0.38–0.43 by 0.40–0.41; sucker width ratio 1:0.83–

0.98. Testes and ovary gathering at junction between middle and posterior thirds of hindbody or slightly posterior to it (Figs. 7 and 8), with distance from middle level of ventral sucker to that of ovary occupying 62–69% of hindbody length. Testes elliptical, diagonal, contiguous, intercecal; anterior (either right or left) testis 0.22-0.25 by 0.09-0.13, posterior 0.22-0.28 by 0.11-0.16. Sperm ducts long; common sperm duct very short (Fig. 9). Prostatic sac subglobular, thinwalled, median, between esophagus and ventral sucker (Figs. 7–9), 0.18–0.25 by 0.13–0.17. Seminal vesicle tubular, convoluted. Pars prostatica club-shaped, surrounded by prostatic cells. Ejaculatory duct short, opening on tip of conelike sinus-organ side by side with metraterm. Genital atrium wide, shallow. Genital pore large, slightly anterior to ventral sucker (Figs. 7-9). Ovary



Figs. 7–10. Azygia rhynogobii sp. nov. 7, holotype, entire body, ventral view; 8, paratype, entire body, ventral view; 9, paratype, terminal genitalia, ventral view, 10, paratype, ovarian complex, dorsal view. c, capsule; csd, common sperm duct; cvd, common vitelline duct; ed, ejaculatory duct; gp, genital pore; lc, Laurer's canal; m, metraterm; mg, Mehlis' gland; o, ovary; od, oviduct; ot, ootype; pp, pars prostatica; ps, prostatic sac; sd, sperm duct; so, sinus-organ; sv, seminal vesicle; u, uterus; vd, vitelline duct. Scale bars: 1 mm in Figs. 7 and 8: 0.2 mm in Figs. 9 and 10.

transversely elliptical, almost level with anterior testis, 0.22-0.26 by 0.12-0.16 (Fig. 10). Ovarian complex anterodorsal to ovary (Fig. 10). Laurer's canal long, opening dorsally through single pore. Seminal receptacle absent. Thin-walled capsule containing distal part of oviduct, common vitelline duct, ootype surrounded by Mehlis' gland, proximal part of uterus, and rarely proximal part of Laurer's canal. Uterus forming close transverse coils in intercecal field between capsule and ventral sucker, serving as uterine seminal receptacle; metraterm well developed, anterior to ventral sucker (Fig. 9). Eggs numerous, 37-64 by 19-38 μm (combined, collapsed): fully-embryonated eggs in paratype from T. hakonensis 56-64 by  $30-38 \mu m$ , and unembryonated ones in holotype and paratype from Rhinogobius sp. 37-56 by 19-30 µm. Vitelline follicles small, extending in extracecal fields from level of posterior margin of ventral sucker or slightly posterior to it to cecal ends. Excretory vesicle bifurcating between anterior testis and ovary (Figs. 7 and 8); arms long, not united anteriorly.

Immature specimens. The morphology and measurements of the 13 immature vouchers (Fig. 11) were as follows: body 1.02–2.13 by 0.43–0.82; forebody 0.53–1.20 long, occupying 44–60% of body length; oral sucker 0.23–0.39 by 0.23–0.40; pharynx 0.07–0.13 by 0.07–0.11; ventral sucker 0.19–0.36 by 0.20–0.40, usually slightly smaller than oral sucker; sucker width sucker width ratio 1:0.76–1.02; testes 0.04–0.14 by 0.04–0.10; and ovary 0.04–0.11 by 0.04–0.15, about equatorial in hindbody, with distance from middle level of ventral sucker to that of ovary occupying 45–57% of hindbody length. Compared with the small body size, the testes and ovary were large and well developed.

Discussion. Azygia rhinogobii sp. nov. is characterized chiefly by (1) a small body, (2) the ventral sucker located about equatorial, (3) the testes and ovary gathering at near the junction between the middle and posterior thirds of the hindbody, (4) the vitelline follicles extending anteriorly close to the posterior margin of the ventral sucker, and (5) the excretory vesicle divided

between the anterior testis and the ovary. In the characteristics (2) to (4), this new species most closely resembles *A. angusticauda* (Stafford, 1904) (syn. *Mimodistomum angusticaudum* Stafford, 1904), which was redescribed by Miller (1941) from North America. *Azygia loossii* Marshall and Gilbert, 1905 and *Ptychogonimus fontanus* Lyster, 1939 also have been synonymized with *A. angusticauda* (see Yamaguti, 1971; Gibson, 1996).

Because no detailed morphological description of A. angusticauda has been available to date, I reexamined the following North American specimens: 2 whole-mounted (1 mature and 1 immature) specimens of M. angusticaudum (syntypes, CMNPA 1900-1654) from Canada (see Stafford, 1904; Miller, 1941); 2 whole-mounted mature specimens of A. angusticauda (CMNPA 1996-0008 and -0009) from Canada (see Miller, 1940); 1 serially-sectioned and 6 whole-mounted mature specimens of P. fontanus (CMNPA 2006-0001 to -0003) from Canada (see Lyster, 1940); 2 serially-sectioned mature specimens of A. loossii (cotypes, USNPC No. 010679.00) from U.S.A. (see Marshall and Gilbert, 1905; Goldberger, 1911); 2 whole-mounted mature specimens of A. angusticauda (USNPC No. 051402.00) from U.S.A. (see Stunkard, 1956, fig. 7); 1 whole-mounted mature specimen of A. angusticauda (USNPC No. 076648.00) from U.S.A. (see Amin, 1982); 1 whole-mounted immature specimen of A. angusticauda (USNPC No. 078925.00) from U.S.A. (Olson, unpublished); 1 whole-mounted immature specimen of A. angusticauda (USNPC No. 081477.00) from U.S.A. (see Aho et al., 1991); 1 whole-mounted immature specimen of A. angusticauda (USNPC No. 089380.03) from Canada (see Bangham, 1941); 3 whole-mounted (1 immature and 2 mature) specimens of A. angusticauda (USNPC No. 095780.00) from U.S.A. (Van Cleave, probably unpublished); and 6 wholemounted (1 immature and 5 mature) specimens of A. angusticauda (USNPC Nos. 095776.00 and 095778.00) from U.S.A. (see Van Cleave and Mueller, 1934).

The mature North American specimens varied

considerably wide in shape and size of the body (oval to slender and 1.44–23.20 long), position of the ventral sucker (the forebody occupying 23-56% of the body length), and in egg size (45–67 by  $21-37 \,\mu m$  (collapsed)). Stafford (1904) and Miller (1941) mentioned nothing about the excretory system in M. angusticaudum. In either Stafford's (1904) syntypes or Miller's (1941) specimens of M. angusticaudum, I could not determine whether the excretory vesicle is bifurcated. In A. loossii, on the other hand, Marshall and Gilbert (1905, fig. 5) and Goldberger (1911, fig. 15) described and figured that the excretory vesicle is roomy behind the posterior testis and that the two longitudinal canals discharge separately into the anterior margin of the vesicle (see also Stunkard, 1956, fig. 6). These conditions were confirmed in Marshall and Gilbert's (1905) cotypes of A. loossii. Lyster (1939) did not describe the excretory vesicle in P. fontanus, either. Lyster's whole-mounted specimens were so highly contracted that I could not trace the excretory vesicle at all. The syntype of *P. fontanus* deposited in The Natural History Museum, London, also was too much contracted (Gibson, 1996; Gibson, personal communication). Lyster's serial sections lacked some of the posterior part of the body including the stem of the excretory vesicle. In the other specimens, the excretory vesicle could not be observed enough to see whether the organ bifurcated. Therefore, it is evident that the excretory vesicle is bifurcated behind the testes at least in A. loossii. Azygia angusticauda needs redescription based on additional, new specimens.

Despite the lack of informations on the excretory vesicle for comparison, the present new species can be distinguished from *A. angusticauda* in having a smaller body (2.30–2.83 long instead of 1.44–23.20 long), smaller oral and ventral suckers, and the ventral sucker located more posterior (the forebody occupying 43–51% of the body length instead of 23–56%). If *A. loossii* is really synonymous with *A. angusticauda*, the present new species is distinct from the latter also in having the excretory vesicle bifurcating between the anterior testis and ovary instead of

behind the testes.

Kakaji (1968) redescribed *A. angusticauda* from India. In Kakaji's specimens measuring 3.33–6.0 mm in body length, the ventral sucker is located about one-third the body length from the anterior end or more anterior to it (figs. 2 and 3), eggs are 30–59 by 28–48  $\mu$ m, and the excretory vesicle is bifurcated behind the posterior testis (fig. 2). Kakaji's species is different not only from *A. angusticauda* but also from the present new species by having the ventral sucker located more anterior and smaller eggs.

In Lake Suwa, Azygia anguillae Ozaki, 1924 [syn. Azygia gotoi (Ariake, 1922)] parasitizes Anguilla japonica Temminck and Schlegel (Anguillidae) (Shimazu, 1979a; this paper). The present new species is readily distinguished from A. anguillae by much smaller body size and much posterior position of the ventral sucker. The adult specimens (2.30-2.83 long) of the present new species are different even from small young adult ones (2.80–5.12 long) of A. anguillae, which appeared to have but recently attained sexual maturity with a few unembryonated uterine eggs (this paper, Fig. 14; see also Shimazu, 1979a, fig. 6), as follows: the ventral sucker is located more posterior (the forebody occupying 43-51% instead of 28-40% of the body length); the testes and ovary gather more compactly and located more posterior (the distance from the middle level of the ventral sucker to that of the ovary occupying 62-69% instead of 46-56% of the hindbody length); and unembryonated uterine eggs are smaller (37–56 by 19–30  $\mu$ m instead of 45– 66 by 27–40  $\mu$ m).

The paratype of the present new species was obtained from the intestine of *Triblolodon hakonensis*, which lacks the stomach. Species of *Azygia* are parasitic in the stomach of fishes in general (Yamaguti, 1971). Possibly, this fish species is not a true final host but an accidental host, which becomes infected with the parasite by ingesting a true final host such as small gobiids.

An unidentified cercaria of the furcocystocercous type has been found in a viviparid snail, Sinotaia quadrata histrica (Gould), from Lake Suwa (Shimazu, 1979a; parthenitae and cercariae obtained in 1973-1975, NSMT-Pl 5310-5313). Because no naturally shed cercariae were available, ten apparently fully-formed cercariae (Figs. 12 and 13) in parthenitae obtained from crashed host snails were measured: body proper 0.91–1.16 by 0.31–0.41; forebody 0.47–0.60 long, occupying 48–55% of body length; oral sucker 0.16-0.23 in diameter; pharynx 0.05-0.06 by 0.04–0.06; ventral sucker usually smaller than oral sucker, 0.13-0.19 by 0.16-0.19, with sucker width ratio of 1:0.84-1.00; testes 0.02-0.04 in diameter; and ovary 0.02–0.05 by 0.02–0.03, with distance from middle level of ventral sucker to ovary 47–58% of hindbody length. The cercaria of A. anguillae, or Cercaria gotoi Ariake, 1922, is also of the furcocystocercous type and develops in another viviparid snail, Cipangopaludina japonica (von Martens) (Shimazu, 1979a). In Lake Suwa, this cercaria has not yet been found although the adult occurs (Shimazu 1979a; this paper). The present cercaria (Fig. 13) is different from that of A. anguillae (see Shimazu, 1979a, fig. 4; this paper) not only in host snail species but also in morphology: the body, oral and ventral suckers, and pharynx are smaller; and the ventral sucker is located more posterior. In morphology, the present cercaria is more similar to the small immature individuals of A. rhinogobii (Fig. 11) than to those of A. anguillae (Fig. 15). It is probable that the present cercaria is the cercarial stage of the present new species, but the exact identification awaits experimental confirmation.

The following immature and mature worms found in the stomach of gobiids from Ibaraki

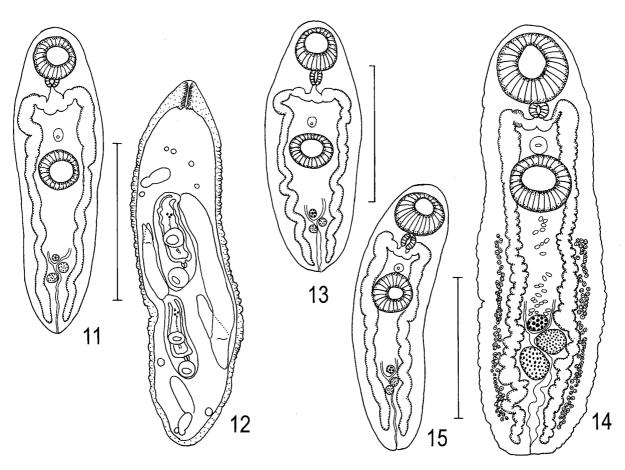


Fig. 11. Azygia rhynogobii sp. nov., immature voucher, ventral view.

Figs. 12 and 13. 12, developing furcocystocercous cercariae in a parthenita, most presumably of *A. rhynogobii*, found in *Sinotaia quadrata histrica* from Lake Suwa; 13, cercarial body proper.

Figs. 14 and 15. Azygia anguillae. 14, small, young adult specimen, ventral view; 15, immature specimen, ventral view. Scale bars: 1 mm in Figs. 11, 12, 14, and 15; 0.5 mm in Fig. 13.

Prefecture, Japan, are also identified as *A. rhynogobii*: from *Tridentiger brevispinis* Katsuyama, Arai and Nakamura from Lake Kitaura at Kitaura in 1994, Gantsu River at Aso in 1994 (NSMT-Pl 5314 and 5315); from *Rhinogobius* sp. from irrigation canals at Itako and Gantsu River, and Lake Kitaura in 1994 (NSMT-Pl 5317–5319); and from *Gymnogobius urotaenia* from the irrigation canals at Itako and Gantsu River in 1994 (NSMT-Pl 5320 and 5321).

#### Azygia anguillae Ozaki, 1924

(Figs. 14 and 15)

Cercaria gotoi Ariake, 1922: 236–238, figs. 1–3, table 1. Azygia anguillae Ozaki, 1924: 426–430, figs. 1–3, fig. 2. Azygia gotoi: Shimazu, 1979a: 229–230, figs. 6–11. (syn. nov.)

Specimens deposited. (1) Immature and mature worms found in the stomach of Anguilla japonica (Anguillidae) from Lake Kizaki in 1981 (NSMT-Pl 5357) and Lake Suwa in 1994 (NSMT-Pl 5360). (2) Immature worms found in the stomach of Rhinogobius sp. (Gobiidae) from Lake Kizaki in 1979 and 1981 (NSMT-Pl 5362 and 5363); Silurus asotus Linnaeus (Siluridae) from Lake Kizaki in 1980, 1981, and 1989 (NSMT-Pl 5364–5366); and Micropterus dolomieu Lacepède (Centrarchidae) from Lake Nojiri at Shinano in 1999 (NSMT-Pl 5367). (3) Fifteen- to 20-day-old immature specimens (NSMT-Pl 5369) of experimental infection in An. japonica made by Shimazu (1979a) and cercariae (NSMT-Pl 5372 and 5373) naturally emerged from Cipangopaludina japonica caught in Lake Kizaki (see Shimazu, 1979a).

Description. 1) Small, young adult specimens (Fig. 14), which appeared to have but recently arrived sexual maturity with a few unembryonated uterine eggs, from *An. japonica* taken in the two lakes were measured: body 2.80–5.12 by 0.62–1.07; forebody 1.07–1.44 long, occupying 28–40% of body length; oral sucker 0.45–0.56 by 0.46–0.51; pharynx 0.13–0.17 by 0.11–0.13; ventral sucker 0.37–0.45 by 0.42–0.50, with sucker width ratio of 1:0.87–0.97; testes 0.16–

- 0.32 by 0.14–0.27; ovary 0.11–0.21 by 0.14–0.21, with distance from middle level of ventral sucker to that of ovary occupying 46–56% of hindbody length; and unembryonated eggs 45–66 by 27–40  $\mu$ m.
- 2) Small, immature specimens from *An. japonica* taken in Lake Kizaki were as follows (Fig. 15): body 0.99–1.92 by 0.35–0.48 long; forebody 0.64–0.80 long, occupying 40–45% of body length; oral sucker 0.25–0.29 by 0.24–0.32; pharynx 0.08–0.11 by 0.06–0.10; ventral sucker 0.22–0.27 by 0.24–0.27, with sucker width ratio of 1:0.80–0.93; testes 0.03–0.08 by 0.03–0.07; and ovary 0.04–0.09 by 0.03–0.08, with distance from middle level of ventral sucker to that of ovary occupying 45–51% of hindbody length.
- 3) Immature specimens from *Rhinogobius* sp., *S. asotus*, and *M. dolomieu* were measured: body 1.44–2.00 by 0.40–0.61; forebody 0.72–0.91 long, occupying 45–51% of body length; oral sucker 0.25–0.30 by 0.25–0.29; pharynx 0.08–0.11 by 0.06–0.09; ventral sucker 0.22–0.27 by 0.22–0.29, with sucker width ratio of 1:0.82–1.00; testes 0.03–0.04 in diameter; and ovary 0.04–0.06 in diameter, with distance from middle level of ventral sucker to that of ovary occupying 46–54% of hindbody length.
- 4) Ten 15- to 20-day-old immature specimens and ten cercariae (in parentheses) were measured: body 1.38–1.51 by 0.48–0.59 (1.10–1.32 by 0.41–0.52); forebody 0.65–0.72 (0.47–0.63) long, occupying 46–50 (47–49)% of body length; oral sucker 0.21–0.25 by 0.21–0.24 (0.21–0.23 in diameter); pharynx 0.09 by 0.07 (0.07–0.09 by 0.06–0.07); ventral sucker 0.19–0.21 by 0.17–0.21 (0.17–0.21 by 0.18–0.21), with sucker width ratio of 1:0.80–0.88 (1:0.86–0.93); testes 0.03 by 0.02–0.03 (0.02–0.03 in diameter); and ovary 0.03–0.04 by 0.02–0.03 (0.04–0.05 in diameter), with distance from middle level of ventral sucker to that of ovary occupying 46–50 (47–59)% of hindbody length.

Previous records from Nagano Prefecture. (1) This species was recorded from the stomach of An. japonica caught in Lakes Kizaki and Suwa (Shimazu, 1979a). (2) The cercaria, or C. gotoi,

was found in *Ci. japonica* taken in Lakes Kizaki and Nakatsuna at Oomachi from 1975 to 1977 (Shimazu, 1979a).

Part of Shimazu's (1979a) specimens were deposited in the NSMT (Shimazu, 1979a), The remainder have been deposited in the NSMT (NSMT-Pl 5358, 5359, 5361, 5368–5373) and in the USNPC (USNPC Nos. 097589.00–097591.00).

Discussion. The morphology of the present specimens from An. japonica agree well with that of A. anguillae as redescribed by Shimazu (1979a).

The measurements of the present immature specimens (1.44-2.00 long) from Rhinogobius sp., S. asotus, and M. dolomieu overlap with those of the immature ones (1.02–2.13 long) of A. rhynogobii (this paper). However, they are identifiable with A. anguillae because the large ones of the former specimens have smaller testes and ovary (0.03-0.04 in diameter instead of 0.04-0.14 by 0.04-0.10; and 0.04-0.06 in diameter instead of 0.04-0.11 by 0.04-0.15, respectively), the ventral sucker located more anterior, and the ovary located more posterior. They more closely resemble the cercariae described above, 15- to 20-day-old immature specimens of experimental infection, and immature ones of natural infection than the immature specimens of A. rhynogobii (this paper).

Most presumably, A. anguillae uses only An. japonica as the final host, in the stomach of which it can reach sexual maturity. Rhinogobius sp. may become infected with unensysted immature worms by ingesting free-living cercariae and then transports them to An. japonica as the second intermediate host or a transport host when it is eaten by An. japonica. Silurus asotus and M. dolomieu may acquire infection of immature worms by ingesting small fish such as Rhinogobius sp. harboring them (see also Shimazu, 1979a).

Small immature worms (NSMT-Pl 5316) were found in *Tridentiger brevispinis* from Lake Kasumigaura at Tamatsukuri in 1994 and 1996. In morphology, they are more similar to *A. anguillae* than *A. rhynogobii* (this paper), but they re-

main unidentified.

Shimazu (1979a) experimentally indicated that *C. gotoi* is the cercaria of *A. anguillae* and proposed a new combination, *A. gotoi* (Ariake, 1922) Shimazu, 1979, for the species name. However, the species name *A. anguillae* is maintained as valid, with *C. gotoi* and *A. gotoi* as its synonyms, in accordance with ICZN Art. 23.7.1 (Anonymous, 1999) because *Cercaria* is a collective-group name (ICZN Art. 67.14).

#### Family Gorgoderidae

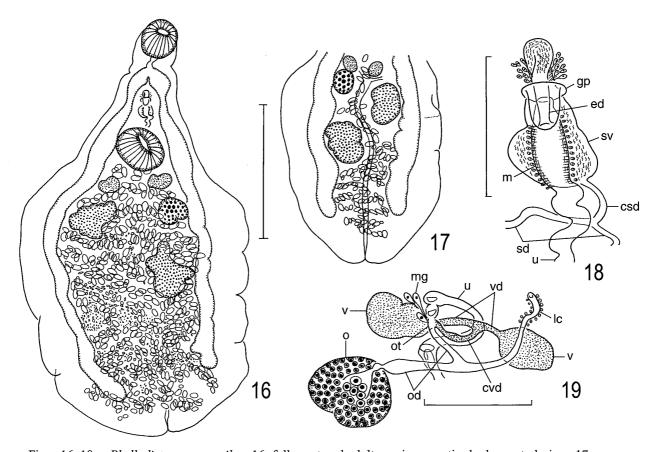
### **Phyllodistomum anguilae** Long and Wai, 1958 (Figs. 16–19)

Phyllodistomum (Phyllodistomum) anguilae Long and Wai, 1958: 351–352, 365–366, fig. 3.

*Phyllodistomum anguilae*: Shimazu, 2005: 142–143, figs. 7–9.

Specimens deposited. Immature and mature worms found in the urinary bladder of *Anguilla japonica* (Anguillidae) from Lake Suwa on 10 September 1976, 30 June 1994, and 9 July 1994 (NSMT-Pl 5322–5325).

Description. Based on 9 well-prepared mature specimens. Body flat, translucent, banjoshaped, 1.47-3.52 by 0.56-1.76; forebody 0.72-1.45 long, occupying 30-53% of body length (Figs. 16 and 17). Tegument smooth. Oral sucker subterminal, 0.15-0.29 by 0.13-0.28. Pharynx absent. Esophagus thick-walled, 0.12-0.34 long, bifurcating at about junction of anterior and middle thirds of forebody; intestinal ceca undulating, ending near posterior extremity of body, with weak diverticula. Ventral sucker usually preequatorial but rarely almost equatorial, 0.18–0.42 by 0.18-0.36; sucker width ratio 1:0.97-1.54. Testes intended irregularly, oblique, separated, intercecal, in second quarter of hindbody; anterior (either right or left) testis 0.14-0.38 by 0.11-0.28, posterior 0.13-0.41 by 0.13-0.38. Sperm ducts long; common sperm duct anterior to ventral sucker, short (Fig. 18). Cirrus pouch absent. Seminal vesicle pyriform, median, dorsal to metraterm, 0.12-0.22 by 0.06-0.12 (Fig. 18). Pars prostatica not seen. Ejaculatory duct long, some-



Figs. 16–19. *Phyllodistomum anguilae.* 16, fully-matured adult specimen, entire body, ventral view; 17, young adult specimen, posterior part of body, ventral view; 18, terminal genitalia, ventral view; 19, ovarian complex, dorsal view. csd, common sperm duct; cvd, common vitelline duct; ed, ejaculatory duct; gp, genital pore; lc, Laurer's canal; m, metraterm; mg, Mehlis' gland; o, ovary; od, oviduct; ot, ootype; sd, sperm duct; sv, seminal vesicle; u, uterus; v, vitellarium; vd, vitelline duct. Scale bars: 1 mm in Figs. 16 and 17; 0.2 mm in Fig. 18; 0.3 mm in Fig. 19.

times everted, distally surrounded by small gland cells, opening into small genital atrium anteriorly to metraterm (Fig. 18). Genital pore median, slightly postbifurcal. Ovary almost globular, sinistro- or dextro-submedian, intercecal, level with anterior testis on the other side of body, 0.10-0.25 in diameter. Ovarian complex median, posterior to ventral sucker (Fig. 19). Mehlis' gland large. Seminal receptacle absent. Oviduct dilated to include spermatozoa. Laurer's canal opening anterodorsally to vitellarium located opposite to ovary. Uterus much folded in hindbody, interand post-cecal; metraterm well developed, anterior to ventral sucker (Figs. 16 and 18); uterine seminal receptacle seen. Uterine eggs numerous, weakly-embryonated eggs 72–83 by 35–41  $\mu$ m, fully-embryonated ones not seen; operculum not seen. Vitellaria in form of 2 compact masses, elliptical or weakly indented, submedian, separated, intercecal, between ventral sucker and ovary, 0.07–0.23 by 0.05–0.12. Excretory vesicle Ishaped, extending anteriorly to level of vitellaria; excretory pore posteroterminal.

Eggs. Eggs were observed in a live mature worm. Eggs measured 43 by  $22 \mu m$  just after formation but 84–96 by 44–52  $\mu m$  in a fully-embryonated stage. The miracidium (or the mother sporocyst) had two flame cells in a formula of 2 [(1)]=2. A daughter sporocyst within the miracidium had four flame cells in a formula of 2 [(1+1)]=4.

Discussion. The present specimens morphologically match the original description of P. (P.) anguilae by Long and Wai (1958) from the urinary bladder of A. japonica and Siniperca chuatsi (Basilewsky) (Percichthyidae) in China and re-

description by Shimazu (2005) from the urinary bladder of *A. japonica* taken in Lake Ogawara at Kamikita, Aomori Prefecture, Japan. Reexamination of Shimazu's specimens (NSMT-Pl 5247) has shown that he measured weakly-embryonated eggs (61–80 by 35–48  $\mu$ m) and that he overlooked fully-embryonated eggs, which were 80–96 by 59–93  $\mu$ m. Long and Wai (1958) measured eggs as 37–89 by 19–40  $\mu$ m in the Chinese specimens. It seems that they measured both unembryonated eggs just after formation and fully-embryonated ones.

This is the first record of this species from Nagano Prefecture.

#### Phyllodistomum mogurndae Yamaguti, 1934

(Figs. 20-22)

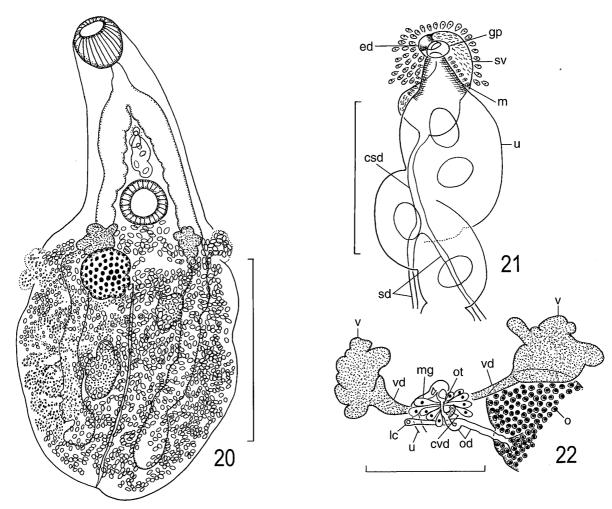
*Phyllodistomum mogurndae* Yamaguti, 1934: 425–428, figs. 87–88.

Specimens deposited. (1) One mature worm each found in the urinary bladder of *Rhinogobius* sp. and *Gymnogobius urotaenia* (Gobiidae) from Lake Suwa on 20 November 1993 and 19 August 1995, respectively (NSMT-Pl 5326 and 5327). (2) The type series of *P. mogurndae* (holotype and 3 paratypes, MPM Coll. No. 22539) found in the urinary bladder of *Odontobutis obscura* (Temminck and Schlegel) [syn. *Mogurnda obscura* (Temminck and Schlegel)] (Odontobutidae) from Lake Ogura, Kyoto Prefecture, Japan, on 20 November 1931 and 4 May 1932 (Yamaguti, 1934).

Description. Based on 2 mature specimens. Body flat, translucent, banjo-shaped, 2.75–5.60 by 1.25–2.40; forebody 1.08–2.48 long, occupying 39–44% of body length (Fig. 20). Tegument smooth. Oral sucker subterminal, 0.28–0.58 by 0.26–0.50. Pharynx absent. Esophagus thickwalled, 0.16–0.50 long, bifurcating at about junction of anterior and middle thirds of forebody; intestinal ceca weakly diverticulated, undulating, ending near posterior extremity of body. Ventral sucker slightly pre-equatorial, 0.25–0.48 by 0.26–0.57; sucker width ratio 1:1.00–1.12.

Testes weakly indented irregularly, oblique, separated, intercecal, in middle third of hindbody; anterior (left) testis 0.22-0.63 by 0.16-0.25, posterior 0.31-0.63 by 0.20-0.50. Sperm ducts long; common sperm duct short, anterior to ventral sucker (Fig. 21). Cirrus pouch absent. Seminal vesicle pyriform, median, dorsal to metraterm, 0.18-0.34 by 0.09-0.14 (Fig. 21). Pars prostatica not seen. Ejaculatory duct short, distally surrounded by small gland cells, opening into small genital atrium anteriorly to metraterm (Fig. 21). Genital pore median, slightly postbifurcal. Ovary slightly indented, dextro-submedian, intercecal, pretesticular, 0.30-0.53 by 0.27-0.40. Ovarian complex internal to ovary (Fig. 22). Oviduct short. Mehlis' gland well developed. Seminal receptacle absent. Laurer's canal short, running transversely to open posteriorly to left vitellarium. Uterus much folded in all available space of hindbody; metraterm well developed, anterior to ventral sucker (Fig. 21); uterine seminal receptacle not seen. Uterine eggs numerous; weakly-embryonated eggs 33–41 by 22–30  $\mu$ m, fully-embryonated 56-64 by  $38-48 \mu m$ ; operculum not seen. Vitellaria in form of 2 compact masses, irregularly indented, symmetrical, ventral to intestinal ceca, between ventral sucker and ovary, 0.19-0.38 by 0.6-0.25. Excretory vesicle Ishaped, extending anteriorly to ovarian level; excretory pore posterodorsal.

Discussion. The present specimens agree with the original description of P. mogurndae by Yamaguti (1934) in morphology, except for the relative size of the two suckers and egg size. Yamaguti described that the ventral sucker is distinctly smaller than the oral sucker. However, the ventral sucker was smaller than the oral sucker in one but larger in the other in the present specimens. Yamaguti measured eggs as 42–48 by 30–  $37 \, \mu \text{m}$ . In the present specimens, weakly- and fully-embryonated eggs were 33-41 by 22-30  $\mu$ m, and 56–64 by 38–48  $\mu$ m, respectively. Examination of the type series revealed that weaklyand fully-embryonated eggs were 25-37 by 17- $27 \mu m$  and 40-48 by  $27-32 \mu m$ , respectively. The difference in size of fully-embryonated eggs be-



Figs. 20–22. *Phyllodistomum mogurndae*. 20, adult specimen, entire body, ventral view; 21, terminal genitalia, ventral view; 22, ovarian complex, dorsal view. Abbreviations as in Figs. 18 and 19. Scale bars: 1 mm in Fig. 20; 0.3 mm in Figs. 21 and 22.

tween Yamaguti's specimens and the present ones is considered to be insignificant because eggs increase in size as embryos in them develop (see also the egg size in *P. anguilae* in this paper).

The following specimens in Yamaguti's Collection are also regarded as *P. mogurndae*: 1 mature specimen (MPM Coll. No. 22540, unidentified, unpublished) found in the urinary bladder of *O. obscura* from Lake Ogura on 9 December 1931; 2 mature specimens (MPM Coll. No. 22019, *P. mogurndae*, unpublished) found in the urinary bladder of *O. obscura* from Obama, Fukui Prefecture, Japan, on 26 March 1935; 3 mature specimens (MPM Coll. No. 22541, *P. mogurndae*, unpublished) found in the urinary bladder of *O. obscura* from Katsura [probably Katsura River in Kyoto Prefecture], Japan, on 15

December 1938; 3 mature specimens (MPM Coll. No. 22268, unidentified, unpublished) from Yodo River (other data not given); 1 mature specimen (MPM Coll. No. 22542, labeled "P. mogurndae (?)", unpublished) found in the urinary bladder of Pseudobagrus nudiceps Sauvage (Bagridae) from Lake Biwa, Shiga Prefecture, Japan, on 7 December 1938; and 1 mature specimen (MPM Coll. No. 22267, unidentified, unpublished) found in the urinary bladder of Ps. nudiceps from Kyoto, Japan, on 29 October 1940. In these specimens, the ventral sucker was not always smaller than the oral sucker, either.

This is the first record of this species from Nagano Prefecture. *Rhinogobius* sp. and *Gymnogobius urotaenia* are new host records for the species (see Shimazu, 2003b).

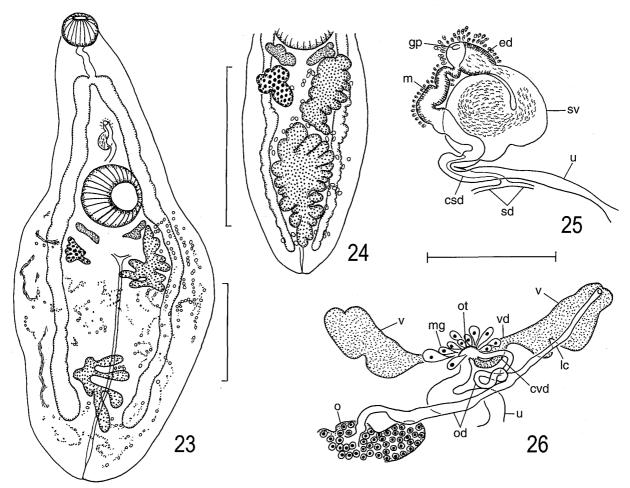
## *Phyllodistomum parasiluri* Yamaguti, 1934 (Figs. 23–26)

Phyllodistomum parasiluri Yamaguti, 1934: 423–425, fig. 86.

Specimens deposited. (1) Immature and mature worms found in the urinary bladder of Silurus asotus Linnaeus (Siluridae) from Lake Kizaki on 5 October 1976, 28 August 1981, and 8 September 1981 (NSMT-Pl 5328–5330); and Lake Suwa on 2 October 1993, 12 May 1994, and 9 June 1994 (NSMT-Pl 5331–5333). (2) The type series of *P. parasiluri* (holotype and 4 paratypes, MPM Coll. No. 22537) found in the urinary bladder of *S. asotus* [syn. *Parasilurus asotus* (Linnaeus)] from Lake Ogura, Kyoto Prefecture, Japan, on 9, [14, and 21] November 1931

#### (Yamaguti, 1934).

Description. Based on 7 mature specimens. Body flat, translucent, lanceolate-oblong or oval, 1.63-4.80 by 0.80-2.40; forebody 0.75-2.67 long, occupying 39-55% of body length (Fig. 23). Tegument smooth. Oral sucker subterminal, 0.17-0.34 by 0.19-0.34. Pharynx absent. Esophagus thick-walled, 0.16-0.34 long, bifurcating at about junction of anterior and middle thirds of forebody; intestinal ceca long, undulating, ending near posterior extremity of body, with weak diverticula. Ventral sucker usually pre-equatorial but rarely almost equatorial, 0.28-0.63 by 0.31-0.60; sucker width ratio 1:1.58-1.97. Testes large, deeply indented irregularly in small, young adult specimens (Fig. 24) but almost digitiform and apparently atrophied in larger, senile adult ones (Fig. 23), oblique, separated, intercecal, in



Figs. 23–26. *Phyllodistomum parasiluri*. 23, senile adult specimen, entire body, ventral view; 24, young adult specimen, hindbody, ventral view; 25, terminal genitalia, ventral view; 26, ovarian complex, dorsal view. Abbreviations as in Figs. 18 and 19. Scale bars: 1 mm in Figs. 23 and 24; 0.3 mm in Figs. 25 and 26.

hindbody; anterior (either right or left) testis 0.25–0.63 by 0.22–0.47, posterior 0.36–0.75 by 0.31-0.57. Sperm ducts long; common sperm duct anterior to ventral sucker, short (Fig. 25). Cirrus pouch absent. Seminal vesicle pyriform, median, dorsal to metraterm (Fig. 25), 0.12-0.28 by 0.03–0.20. Pars prostatica not seen. Ejaculatory duct short, distally surrounded by small gland cells, opening into small genital atrium anteriorly to metraterm (Fig. 25). Genital pore median, midway between intestinal bifurcation and ventral sucker. Ovary lobed, dextro- or sinistro-submedian, intercecal, level with anterior testis on the other side of body, 0.25-0.31 by 0.17-0.25, apparently atrophied in large, senile adult specimens. Ovarian complex median, posterior to ventral sucker (Fig. 26). Oviduct before ootype long, convoluted, including spermatozoa. Mehlis' gland large. Seminal receptacle absent. Laurer's canal opening dorsally to vitellarium located opposite to ovary. Uterus weakly folded and almost intercecal in small, young adult specimens (Fig. 24) but much folded in all available space of hindbody in larger, senile adult ones (Fig. 23); metraterm well developed, anterior to ventral sucker (Fig. 25); uterine seminal receptacle present. Uterine eggs numerous, weakly-embryonated eggs 32–38 by 24–28  $\mu$ m, fully-embryonated ones 40-61 by 34-48  $\mu$ m; operculum not seen. Vitellaria in form of 2 compact masses, transversely elongate, irregularly indented, separated, almost intercecal, between ventral sucker and ovary, 0.15-0.31 by 0.06-0.12. Excretory vesicle I-shaped, extending anteriorly to ovarian level; excretory pore posteroterminal.

Previous record from Nagano Prefecture. Yamaguti had already found three mature specimens of this species in *S. asotus* from Lake Suwa in 1935, but he did not describe them, as mentioned below.

Discussion. The present specimens morphologically agree with Yamaguti's (1934) original description of *P. parasiluri* and the type series.

The following specimens, which were obtained from the urinary bladder of *S. asotus*, in Yamaguti's Collection are also identified as *P. parasil*-

uri: 1 immature specimen (MPM Coll. No. 22538, unidentified, unpublished) from Lake Biwa on 1 November 1931; 2 mature specimens (MPM Coll. No. 22018, P. parasiluri, unpublished) from Okinohata, Fukuoka Prefecture, Japan, on 23 April 1935; 3 mature specimens (MPM Coll. No. 22017, P. parasiluri, unpublished) from Lake Suwa on 16 May 1935; 1 mature specimen (MPM Coll. No. 22263, P. parasiluri, unpublished) from Yodo [probably the Yodo River] on 12 December 1939; and 1 mature specimen (MPM Coll. No. 22266, unidentified, unpublished) from Kyoto on 2 November 1940. The present specimens as well as Yamaguti's ones suggest that, as adult worms grow older, the testes and ovary become atrophied.

Lake Kizaki is a new locality record for the species.

# Family Lissorchiidae Anapalaeorchis hamajimai Fujino and Kifune, 1991

Cercaria monostyloides Ito, 1960: 68–69, fig. 15. (syn. nov.)

Anapalaeorchis hamajimai Fujino and Kifune, 1991: 35–36, figs. 1–8; Shimazu, 1992: 12, 14, figs. 12–16.

Specimens deposited. Immature and mature worms found in the intestine of *Cobitis biwae* Jordan and Snyder (Cobitidae) from Metoba River at Hara, Matsumoto, in 1993, 1994, and 1999 (NSMT-Pl 5493–5495).

Discussion. The present specimens morphologically agree with the original description of *A. hamajimai* by Fujino and Kifune (1991) and redescription by Shimazu (1992).

At the same sampling site in the Metoba River, a tailless cercaria (NSMT-Pl 5496–5500) was obtained from the snail *Semisulcospira libertina* in 1992–1995 and 2000. This cercaria was regarded as *Cercaria monotyloides* described by Ito (1960) from *S. libertina* in Shizuoka Prefecture, Japan, because of their morphological concordance. As seen in the adult of *A. hamajimai*, the cercaria already possessed a pair of slightly diagonal testes (see Shimazu, 1992), although Ito did not de-

scribe it. Furthermore, no lissorchiid species other than *A. hamajimai* has been found in fishes from this river (this paper). From circumstantial evidence, I conclude that *C. monostyloides* is the cercarial stage of *A. hamajimai*, as suggested by Shimazu (1992, 2003b).

This is the first record of this species from Nagano Prefecture.

### Asymphylodora macrostoma Ozaki, 1925 (Figs. 27–30)

Cercaria H: Kobayashi, 1918: 70–73, fig. 16. (syn. nov.)
Cercariaeum A: Kobayashi, 1922: 266–267. (syn. nov.)
Cercariaeum innominatum: Faust, 1924: 295. (syn. nov.)
Asymphylodora macrostoma Ozaki, 1925: 104–106, fig. 4; Shimazu, 1992: 8–10, figs. 6–11.

Parasymphylodora macrostoma: Szidat, 1943: 44–45, fig. 12.

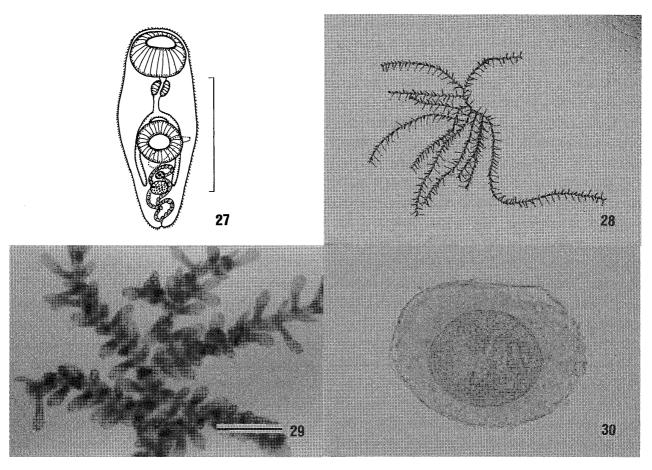
Cercaria innominatum: Ito, Mochizuki and Noguchi, 1959: 918. (syn. nov.)

Orientotrema macrostoma: Tang, 1962: 169, 182, pl. 1, fig. 2.

Specimens deposited. Immature and mature worms found in the intestine of *Tribolodon hakonensis* (Cyprinidae) from Torii River on 7 May 1994 and 11 November 1995 (NSMT-Pl 5334 and 5335); Lake Suwa on 14 September 1992 (NSMT-Pl 5336); and Hiroi River in 1999 and 2004 (NSMT-Pl 5337–5341; USNPC No. 097592.00).

*Life cycle.* Results obtained in the life cycle study are briefly summarized as follows.

1) Adults were found in the intestine of *T. hakonensis* from the Torii and Hiroi rivers (Shimazu, 1992; this paper). On one occasion, two mature worms (NSMT-Pl 5335) were found in a



Figs. 27–30. Asymphylodora macrostoma. 27, cercaria (Cercaria innominata) found in Semisulcospira libertina, entire body, ventral view; 28, an aggregation of cercariae (scale unknown), naturally shed; 29, another aggregation of cercariae, enlarged, naturally shed; 30, encysted metacercaria (cyst 250 by 211 μm; cyst wall 2 μm thick) found in Pseudorasbora parva 14 days after experimental infection, surrounded by host's tissue (429 by 312 μm). Scale bars: 0.5 mm in Fig. 29; 0.2 mm in Fig. 27.

fish as small as 35 mm in standard body length from the Torii River on 11 November 1995.

- 2) Metacercariae were found encysted in the connective tissue of the mucous membrane chiefly of the gill arches and gill rakers, rarely of the oral cavity and pharynx, and more rarely of the intestine of cyprinids, *T. hakonensis*, *Gnathopogon elongatus* (Temminck and Schlegel), *Moroco steindachneri* (Sauvage), *Pseudorasbora parva* (Temminck and Schlegel), and *Zacco platypus* (Temminck and Schlegel), from the Hiroi River in 1995–2004 (NSMT-Pl 5342 and 5343); and *T. hakonensis* from Lake Suwa on 18 September 1999 (NSMT-Pl 5344).
- 3) Daughter rediae and cercariae (NSMT-Pl 5345–5347) were found in snails, *Semisulcospira libertina* from the Torii River in 1993–1995 and *S. libertina* and *S. dolorosa* from the Hiroi River in 1995–1999.
- 4) Cercariae (Fig. 27) usually became aggregated in the mantle cavity of host snails when they were leaving the hosts (Figs. 28 and 29). Aggregations were attained as follows: a second cercaria stuck with the ventral sucker to a first at the back exactly dorsal to the ventral sucker; the two cercariae made at right angles to each other; similarly, a third cercaria stuck to the second; and, thus, a chain of cercariae was formed, making a clockwise spiral. Sometimes, two cercariae stuck to one cercaria, one to the back and the other to the posterior tip of the body; then one cercaria each stuck to the back of the respective cercariae; and, thus, the chain became branched here. The aggregation was flesh-colored and moved slowly on the bottom of the water.
- 5) When cercariae were experimentally exposed to small fish (*T. hakonensis* and *P. parva*), they were easily ingested by the fish and eventually found encysted in the connective tissue of the mucous membrane chiefly of the gill arches and gill rakers and rarely of the oral cavity and pharynx and more rarely of the intestine of the fish (Fig. 30, NSMT-Pl 5348 and 5349). On day 7 to 28 after infection, some were found encysted in the gills, and some others were found unencysted and free in the intestinal lumen (NSMT-Pl

- 5351–5354). The latter grew slightly further than the encysted metacercariae but were still immature. When cercariae were injected with a stomach tube into the intestine of *T. hakonensis*, 3 days later some metacercariae were found encysted in the intestinal wall, but some others were found unencysted and free in the intestinal lumen (NSMT-PI 5350).
- 6) When encysted metacercariae obtained from experimentally infected *P. parva* 14 or 15 days after infection were fed to *T. hakonensis*, immature and weakly mature worms were recovered from the intestine 14 days later (NSMT-Pl 5355), and fully mature ones were recovered 18 and 42 days later (NSMT-Pl 5356).

Previous records from Nagano Prefecture. This species was recorded from the intestine of *T. hakonensis* caught in Torii and Nogu rivers and Lake Suwa (Shimazu, 1992).

Discussion. The present adult specimens of both natural and experimental infections in *T. hakonensis* are identified as *A. macrostoma* because they morphologically agree with the original description of *A. macrostoma* by Ozaki (1925) as well as redescription by Shimazu (1992). The Hiroi River is a new locality record for the species.

The present metacercariae of both natural and experimental infections also are identified as A. macrostoma because they morphologically agree with those of A. macrostoma described by Yamaguti (1934, 1938) and Okabe (1940) from several species of freshwater fishes in Japan (see Shimazu, 1992) and the present immature specimens of A. macrostoma of natural infection in T. hakonensis. The present daughter redia and cercaria are regarded as Cercariaeum innominatum, or Cercaria innominata (originally spelt innominatum), because they are morphologically identical with C. innominata, which was redescribed by Ito (1960) from S. libertina in Shizuoka Prefecture, Japan. I conclude that C. innominata is the cercarial stage of A. macrostoma.

The above-mentioned results outline the life cycle of *A. macrostoma* (see also Shimazu, 1997). The snails *S. libertina* and *S. dolorosa* 

serve as the first intermediate host. Cyprinids act as the second intermediate host. The encystment is necessary to metacercariae in it. Shimazu (1992) listed other second intermediate and final host fishes known at that time. The fish *T. hakonensis* serves as the final host.

Tang (1962) obtained cercariae from the snail *Melania peregrinorum* Heude (Pleuroceridae) in China; fed them to a cyprinid fish, *Puntia* sp.; and, 15 days later, recovered adults identifiable to *A. macrostoma* (syn. *Orientotrema macrostoma*) from the fish. The worms reached sexual maturity slightly earlier than those of the present results. Tang mentioned nothing about the encysted metacercarial stage.

From the present results, I consider that T. hakonensis (the final host) becomes infected with A. macrostoma by ingesting small cyprinids (the second intermediate host) harboring encysted metacercariae. As mentioned above, T. hakonensis as small as 35 mm in standard body length was found already infected with fully mature adults. Such a small fish seems unlikely to eat fish infected with metacercariae. When cercariae were injected into T. hakonensis, encysted metacercariae were found in the intestinal wall, and unencysted worms were found free in the intestinal lumen 7 to 28 days after infection. The latter worms grew slightly further than the encysted metacercariae but were still immature. It is possible that, after liberating from their cysts in the intestinal wall into the intestinal lumen, by unknown mechanism after unknown days after infection, now juveniles can attain sexual maturity there. This needs experimental confirmation.

Cercariae became aggregated when leaving snail hosts. Okumura (1919) briefly described a similar aggregation of tailless cercariae obtained from *S. libertina* in Okayama, Japan. Okumura also must have had the aggregation of cercariae of *A. macrostoma*. I believe that, by moving slowly like some invertebrates on the bottom of the water, flesh-colored aggregations of cercariae attract bottom-feeding fishes of the second intermediate host, and allow the fishes to ingest them not only much more easily but also much more

efficiently than a single cercaria does. Beuret and Pearson (1994) discussed some other such cercarial aggregations.

#### Palaeorchis diplorchis (Yamaguti, 1936)

Asymphylodora diplorchis Yamaguti, 1936: 4–5, fig. 8. Steganoderma kamatukae Takeuti, 1936: 581–583, 1 fig. Palaeorchis diplorchis: Szidat, 1943: 48, fig. 14; Shimazu, 1992: 15, 17, figs. 17–22.

Previous record from Nagano Prefecture. This species was recorded as A. diplorchis by Yamaguti (1936) from the intestine of Pseudogobio esocinus (Temminck and Schlegel) (Cyprinidae) (type host) from Lake Suwa (type locality) (see also Shimazu, 1992).

Discussion. I examined 18 specimens of *P. esocinus* from Lake Suwa from 1991 to 1999, but not a single specimen of the species was obtained from them.

# Family Didymozoidae *Philopinna higai* Yamaguti, 1936

Philopinna higai Yamaguti, 1936: 1-2, figs. 1-7.

Specimens deposited. (1) Immature and mature worms found in the fins of Sarcocheilichthys variegatus microoculus Mori (Cyprinidae) from Lake Kizaki in 1980, 1981, and 1983 (NSMT-Pl 5374-5376); Lake Suwa in 1991-1993 (NSMT-Pl 5377-5380); and Hiroi River in 1995, 1999, and 2000 (NSMT-Pl 5381-5387; USNPC No. 097594.00). (2) The type series of P. higai (MPM Coll. No. 22297) from the fins and orbits of "Sarcocheilichthys variegatus (Temm. et Schleg.)" (Cyprinidae) (type host) from Lake Suwa (type locality): holotype on [31 March 1936] (not 18 May 1935); and many paratypes on [12 June 1932, 31 March 1935, 8 and 18 May 1935, 30 and 31 March 1936, and 12 June 1936] (Yamaguti, 1936).

Previous record from Nagano Prefecture. As mentioned above, this species was found in "Sarcocheilichthys variegatus" from Lake Suwa (Yamaguti, 1936).

Discussion. These specimens morphologi-

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cally agree with Yamaguti's (1936) original description of *P. higai* and the type series. It is possible that the type host is *S. v. microoculus* because it is only this subspecies in the genus that lives in Lake Suwa at present.

The following hosts and localities for P. higai are recorded for the first time, although they are not collected from Nagano Prefecture: (1) S. v. microoculus from Kitaura at Kitaura, Ibaraki Prefecture, Japan, in 1994 (NSMT-Pl 5388); Lake Biwa at Ono'e, Kohoku, and at Minamihama, Shiga, Shiga Prefecture, Japan, in 1976 and 1982, respectively (NSMT-Pl 5389 and 5390); and Uji River at Uji, Kyoto Prefecture, Japan, in 1979 (NSMT-Pl 5391); (2) S. biwaensis Hosoya from Lake Biwa at Minamihama in 1982 (NSMT-Pl 5392); (3) S. v. variegatus (Temminck and Schlegel) from the Yoshii River at Joto, Okayama, Okayama Prefecture, Japan, in 1976 (NSMT-Pl 5393); Ashida River at Honjo, Fukuyama, Hiroshima Prefecture, Japan, in 1980 (NSMT-Pl 5394); Yanagawa, Fukuoka Prefecture, Japan, in 1976 (NSMT-Pl 5395); and Midori River at Kosa, Kumamoto Prefecture, Japan, in 1977 (NSMT-Pl 5396); and (4) S. v. wakiyae Mori from Puk'an-gang River at Anbo-ri, Somyon, Chunsong-gun, Kangwon-do, Korea, in 1984 (NSMT-Pl 5397). Except for the first from Kitaura, the others were dissected out of the fishes collected by Prof. K. Hosoya (Kinki University, Nara).

# Family Derogenidae Genarchopsis fellicola Shimazu, 1995

Genarchopsis goppo: Yamaguti, 1938: 133, in part, not of Ozaki, 1925.

Genarchopsis fellicola Shimazu, 1995: 13–14, figs. 7–11.

Specimens deposited. Adult worms found in the gall bladder of *Gymnogobius urotaenia* (syn. *Chaenogobius urotaenia*) (Gobiidae) from Lake Suwa in 1998 and 1999 (NSMT-Pl 5398; USNPC No. 097597.00).

Previous records from Nagano Prefecture. This species was recorded from the gall bladder of Gy. urotaenia (type host) and Rhinogobius

brunneus (Rutter) (=Rhinogobius sp.) (Gobiidae) caught in Lake Suwa (type locality) (Yamaguti, 1938; Shimazu, 1995).

Discussion. The present specimens morphologically agree with Shimazu's (1995) original description of *G. fellicola*.

#### Genarchopsis goppo Ozaki, 1925

*Genarchopsis goppo* Ozaki, 1925: 101–103, figs. 1–3; Shimazu and Urabe, 2005: 2–3, figs. 1–3.

Progonus goppo: Srivastava, 1933: 55.

Genarchopsis anguillae Yamaguti, 1938: 132–133, fig. 81.

Genarchopsis gigi Yamaguti, 1939: 227, pl. 29, fig. 6.

Genarches anguillae: Skryabin and Gushanskaya, 1955: 678, 680, fig. 199.

*Genarches gigi*: Skryabin and Gushanskaya, 1955: 678, 680, 685, fig. 200.

Genarches goppo: Skryabin and Gushanskaya, 1955: 678, 685–686, 689, fig. 201.

Genarchapsis [sic] goppo: Shimazu, 1995: 6-9, figs. 1-4.

Specimens deposited. Immature and mature worms obtained in Lake Suwa as follows: from the stomach of *Gymnogobius urotaenia* (syn. *Chaenogobius urotaenia*) (Gobiidae) in 1994, 1996, 1998, and 1999 (NSMT-PI 5399–5404) and *Rhynogobius* sp. (Gobiidae) in 1999 (NSMT-PI 5405; USNPC No. 097596.00); and from the stomach and intestine of *Cottus pollux* Günther (Cottidae) in 1999 (NSMT-PI 5406).

Previous records from Nagano Prefecture. This species was recorded from the stomach of Rhinogobius brunneus (=Rhinogobius sp.) and Micropterus salmoides Lacepède (Centrarchidae) caught in Lake Kizaki; and Gy. castaneus (O'Shaughnessy) [syn. C. laevis (Steindachner)], Gy. urotaenia, Rhinogobius sp., Silurus asotus (Siluridae), Anguilla japonica (Anguillidae), and M. salmoides caught in Lake Suwa (Shimazu, 1995).

Discussion. The present specimens morphologically agree with the original description of *G. goppo* by Ozaki (1925) and redescription by Shimazu (1995). Shimazu and Urabe (2005) regarded *G. anguillae* as a synonym of *G. goppo*. Urabe (2001) experimentally showed that a cystophorous cercaria produced in a redia in the snail *Semisul*-

cospira libertina in Nara developed into an adult identifiable to *G. goppo*. This cercaria morphologically resembles *Cercaria yoshidae* Cort and Nichols, 1920 (Urabe, 2001). Similar cercariae have been found in *S. libertina* from Hoshina, Wakaho, Nagano (Shimazu and Shimizu, 1984); and *S. reiniana* from Lake Suwa (my unpublished data).

# Family Isoparorchiidae *Isoparorchis hypselobagri* (Billet, 1898)

*Distomum hypselobagri* Billet, 1898: 283, 288–290, pl. 13, fig. 8.

*Isoparorchis trisimilitubis* Southwell, 1913: 92–94, pls. 8–9, figs. 9–12.

Leptolecithum eurytremum Kobayashi, 1915: 50–52, pl. 2, fig. 1; Kobayashi, 1921: 397–399, pl. 26, fig. 1.

Isoparorchis eurytrema: Travassos, 1922: 230.

*Isoparorchis tandani* Johnston, 1927: 129, 131–132, text fig. A, figs. 1–4.

Isoparorchis hypselobagri: Ejsmont, 1932: 456.

Specimens deposited. Mature worms found in the air bladder of Silurus asotus (Siluridae) from Lake Kizaki in 1980 (NSMT-Pl 5407) and Lake Suwa in 1992 and 1993 (NSMT-Pl 5408 and 5409).

Previous records from Nagano Prefecture. This species (immature worms or juveniles) was recorded as *I. trisimilitubis* from the body cavity of Gnathopogon elongatus [syn. G. e. elongatus (Temminck and Schlegel)] (Cyprinidae) and "Chaenogobius macrognathos (Bleeker)" (Gobiidae) caught in Lake Suwa (Yamaguti, 1938).

Discussion. The present specimens were morphologically similar to the specimens that Yamaguti (1934) described as L. trisimilitubis from the air bladder of S. asotus (syn. Parasilurus asotus) (3 immature and 2 mature specimens, MPM Coll. No. 22009, from Lake Biwa on 9 July 1927); and a mature specimen (MPM Coll. No. 22013) in Yamaguti's Collection. The latter is labeled "Isoparorchis hypserobagri (Billet, 1898), Host: Tandanus tandanus, 12–25". Because Yamaguti (1934) mentioned nothing about this specimen, it is not known that the specimen is one of the specimens that Johnston (1927) de-

scribed as *I. tandani* from *T. tandanus* in Australia.

Yamaguti's Collection includes nine juveniles of I. trisimilitubis from Lake Suwa (Yamaguti, 1938): eight from G. e. elongatus (Japanese name "tamoroko") (MPM Coll. No. 22007, on 17 May 1935 and 31 March 1936) and one from "Chaenogobius macrognathos" (MPM Coll. No. 22008, on 31 March 1936). The latter host is labeled "ukigori" in Japanese on the slide. The scientific name of "ukigori" is Gymnogobius urataenia. Shimazu (2003b) made the mistake of guessing that "Chaenogobius macrognathos" should be Gy. castaneus (O'Shaughnessy) [syn. C. laevis (Steindachner), not Gy. breunigii (Steindachner)]. Yamaguti's Collection also includes a juvenile of "Isoparorchis" (MPM Coll. No. 22022) from the body cavity of "moroko" in Japanese from Lake Suwa. The Japanese host name "tamoroko" written on the label of the slide (MPM Coll. 22007) suggests that this "moroko" should be read "tamoroko" or G. e. elongatus.

This is the first record of the adult of this species in Nagano Prefecture.

## Family Allocreadiidae Allocreadium aburahaya Shimazu, 2003

*Allocreadium aburahaya* Shimazu, 2003a: 121–122, figs. 4–6.

Previous record from Nagano Prefecture. This species was recorded from the intestine of Moroco steindachneri (Sauvage) (syn. Phoxinus lagowskii steindachneri Sauvage) (type host) caught in Hiroi River (type locality) (Shimazu, 2003a).

#### Allocreadium gotoi (Hasegawa and Ozaki, 1926)

Macrolecithus gotoi Hasegawa and Ozaki, 1926: 225–227, fig. 1, 1 table.

Allocreadium gotoi: Shimazu, 1988a: 6-7, figs. 1-3.

Previous records from Nagano Prefecture. This species was recorded from the intestine of Misgurnus anguillicaudatus (Cantor) (Cobitidae) caught in Komi most presumably in Nagano Pre-

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fecture (Yamaguti, 1934; Shimazu, 1988a); Lake Kizaki (Shimazu, 1988a); a small river at Okada, Matsumoto; a small river at Midori, Iiyama; Lake Suwa; and Furukawa River at Toyota (Shimazu, 2002).

Discussion. Shimazu (2002) obtained a cercaria of the ophthalmoxiphidiocercous type from *Pisidium nikkoense* Mori (Bivalvia, Pisidiidae) collected in the river at Midori, Iiyama, and regarded it as the cercarial stage of *Allocreadium gotoi*. The entire life cycle of this species is still unknown.

#### Allocreadium shinanoense Shimazu, 2003

Allocreadium shinanoense Shimazu, 2003a: 119–120, figs. 1–3.

Previous record from Nagano Prefecture. This species was recorded from the intestine of Moroco steindachneri (syn. Phoxinus lagowskii steindachneri) (Cyprinidae) (type host) caught in Hiroi River (type locality) (Shimazu, 2003a).

#### Family Opecoelidae

#### Dimerosaccus oncorhynchi (Eguchi, 1931)

Allocreadium oncorhynchi Eguchi, 1931: 21–22; Eguchi, 1932: 24–28, 1 pl., figs. 1–6.

Plagioporus oncorhynchi: Peters, 1957: 140.

Dimerosaccus oncorhynchi: Shimazu, 1980: 164, 166, figs. 1–7; Shimazu, 1988b: 10–11, figs. 5–7; Shimazu, 2000: 25–26, figs. 11–13; Shimazu and Urabe, 2005: 4–5, figs. 4–7.

*Plagioporus honshuensis* Moravec and Nagasawa, 1998: 283–284, fig. 1.

Specimens deposited. Immature and mature worms found in the intestine of Salvelinus leucomaenis pluvius (Hilgendorf) (Salmonidae) from Ide River at Araya, Iiyama, in 1995 (NSMT-Pl 5463 and 5464).

Description. The vitelline follicles did not reach anteriorly to the bifurcal level in some of the present specimens but entered the prebifurcal region of the body in others.

Previous records from Nagano Prefecture. This species was recorded from the intestine of S.

*l. pluvius* from Samu River at Fujisawa, Iiyama (Shimazu, 1980); *S. l. pluvius* and *Cottus pollux* (Cottidae) from Ide River at Araya, Iiyama (Shimazu, 2000); and *S. l. pluvius* from Hime and Matsu rivers and Nakakurozawa (a small river) at Hakuba (Shimazu, 1988b, 2000).

Discussion. Two morphological forms have been known in this species from Japan: Honshu form, in which the anterior limit of the vitelline follicles is posterior to the bifurcal level; and Hokkaido form, in which the limit is anterior (Shimazu, 2000). Shimazu and Urabe (2005) considered this difference in anterior limit of the vitelline follicles a morphological intraspecific variation in the species because they observed both forms in the material collected in Nara Prefecture. Both forms also were observed in the present specimens, which supports Shimazu and Urabe (2005).

### Neoplagioporus elongatus (Goto and Ozaki, 1930)

Lebouria elongata Goto and Ozaki, 1930: 75–76, fig. 2. Caudotestis orientalis Yamaguti, 1934: 288–290, fig. 19. Caudotestis gnathopogonis Yamaguti, 1934: 290–292, fig. 20.

Plagioporus elongata [sic]: Price, 1934: 6.

Plagioporus (Caudotestis) elongatus: Yamaguti, 1954: 76. Plagioporus (Caudotestis) gnathopogonis: Yamaguti, 1954: 76.

Plagioporus (Caudotestis) orientalis: Yamaguti, 1954: 76.Plagioporus (Plagioporus) elongatus: Skryabin and Koval', 1958: 459–460, fig. 148.

Plagioporus (Plagioporus) orientalis: Skryabin and Koval', 1958: 494–498, fig. 163.

Plagioporus orientalis: Koval', 1959: 129.

Neolebouria elongatus: Gibson, 1976: 252.

Neoplagioporus elongatus: Shimazu, 1990b: 393-394, figs. 10-17.

Specimens deposited. Immature and mature worms found in the intestine of Sarcocheilichthys variegatus microoculus (Cyprinidae) in 1991 and 1993 (NSMT-Pl 5465–5469); Pseudorasbora parva (Cyprinidae) in 1991, 1993, 1994, and 1999 (NSMT-Pl 5470–5473); Pseudogobio esocinus (Cyprinidae) in 1991, 1992, and 1999 (NSMT-Pl 5474–5476); Gnathopogon elongatus

(Cyprinidae) in 1992 (NSMT-Pl 5477); Hemibarbus barbus (Temminck and Schlegel) (Cyprinidae) in 1999 (NSMT-Pl 5478); Carassius auratus subsp. (Japanese name "nagabuna") (Cyprinidae) in 1992 (NSMT-Pl 5479); and Gymnogobius urotaenia (Gobiidae) in 1993 (NSMT-Pl 5480) all from Lake Suwa.

Previous record from Nagano Prefecture. This species was recorded from the intestine of S. variegatus [probably S. v. microoculus] from Lake Suwa (Shimazu, 1990b).

Discussion. Pseudorasbora. parva, Ps. esocinus, G. elongatus, H. barbus, C. auratus subsp., and Gy. urotaenia are new host records for the species from Lake Suwa. Pseudorasbora parva and C. auratus subsp. are new host records from Japan (see Shimazu, 1990b, 2003b; Shimazu and Urabe, 2005).

#### Neoplagioporus zacconis (Yamaguti, 1934)

Caudotestis zacconis Yamaguti, 1934: 292–294, fig. 21. Plagioporus (Caudotestis) zacconis: Yamaguti, 1954: 76. Plagioporus (Plagioporus) zacconis: Skryabin and Koval', 1958: 533–534, 537, fig. 180.

Neoplagioporus zacconis: Shimazu, 1990b: 387–388, figs. 1–5.

Specimens deposited. Immature and mature worms found in the intestine of Zacco platypus (Cyprinidae) from Lake Suwa in 1991 (NSMT-Pl 5482), Hiroi River in 1996 and 1999 (NSMT-Pl 5483–5487; USNPC No. 097598.00), and Tenryu River in 2000 (NSMT-Pl 5488).

Previous records from Nagano Prefecture. This species was recorded from the intestine of Z. platypus from Lake Suwa (Yamaguti, 1938, 1942) and Chikuma River at Ueda (Shimazu, 1990b).

*Discussion*. The Hiroi and Tenryu rivers are new locality records for this species.

#### Urorchis acheilognathi Yamaguti, 1934

*Urorchis acheilognathi* Yamaguti, 1934: 415–417, figs. 81–82; Shimazu, 1990a: 208–209, figs. 9–15.

Specimens deposited. A mature worm found

in the intestine of *Pseudorasbora parva* (Cyprinidae) from Lake Suwa in 1991 (NSMT-Pl 5470).

Previous records from Nagano Prefecture. This species was recorded from the intestine of Tanakia lanceolata (Temminck and Schlegel), P. parva, Moroco steindachneri, Sarcocheilichthys variegatus microoculus (Cyprinidae), and "kizakimasu" [a lake form of Oncorhynchus masou masou (Brevoort), or this form  $\times$  O. rhodurus Jordan and McGregor (Japanese name "biwamasu"), or both] (Salmonidae) from Lake Kizaki (Shimazu, 1990a).

Discussion. Lake Suwa is a new locality record for this species in Nagano Prefecture.

#### Urorchis goro Ozaki, 1927

*Urorchis goro* Ozaki, 1927: 160–163, figs. 5–7; Shimazu, 1990a: 205–207, figs. 1–8.

Specimens deposited. Immature and mature worms were found in the intestine of *Rhinogobius* sp. (Gobiidae) from Lake Suwa in 1993 (NSMT-Pl 5301), a small stream at Oomura, Matsumoto, in 1995 (NSMT-Pl 5489), and Hiroi River in 1999 (NSMT-Pl 5490); and *Cottus pollux* (Cottidae) from Lake Suwa in 1996 (NSMT-Pl 5491).

Previous records from Nagano Prefecture. This species was recorded from the intestine of Lefua echigonia Jordan and Richardson (Balitoridae) from Midori, Iiyama; Rhinogobius brunneus (=Rhinogobius sp.) (Gobiidae) from Matsuoka, Matsumoto; and Gnathopogon elongatus (Cyprinidae) from Lake Suwa (Shimazu, 1990a).

Discussion. Cottus pollux is a new host record for this species in Japan (see Shimazu, 1990a, 2003b). The Hiroi River is a new locality record in Nagano Prefecture.

# Family Heterophyidae \*Pseudexorchis major\* (Hasegawa, 1935)

(Figs. 31-34)

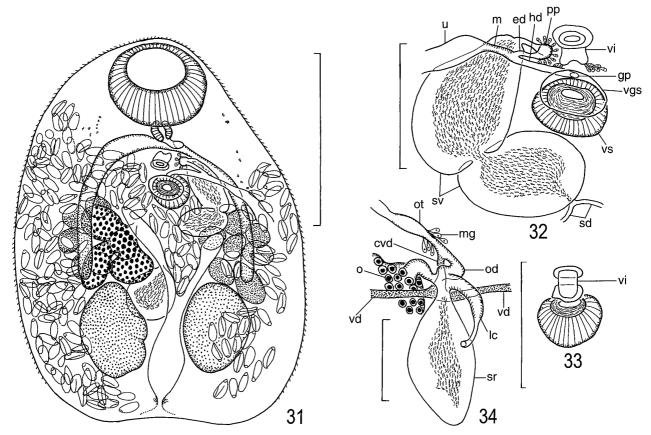
Exorchis major Hasegawa, 1935a: 1192–1197, 1 pl., figs. 1–2.

Pseudexorchis major: Yamaguti, 1938: 66, 68.

Specimens deposited. Immature and mature worms found in the intestine of Silurus asotus (Siluridae) from Lake Kizaki in 1976, 1980, 1981, and 1983 (NSMT-Pl 5502–5507); Nogu River in 1987 (NSMT-Pl 5508); Lake Suwa in 1975, 1991, and 1996 (NSMT-Pl 5509 and 5510; USNPC No. 097593.00); Hiroi River in 1996 (NSMT-Pl 5511); and a small river at Koshoku in 1990 (NSMT-Pl 5512).

Description. Based on 10 fully-matured adult specimens. Body oval to elliptical, 0.60–0.66 by 0.38–0.52; forebody 0.21–0.24 long, occupying 35–39% of body length (Fig. 31). Tegumental scales dense, rotundate on anterior parts of body, becoming sparse acute spines posteriorly, not seen on posteriormost part of body. Eyespot pig-

ment solid or dispersed. Oral sucker subterminal, round, large, 0.12–0.15 by 0.13–0.16. Prepharynx very short. Pharynx elliptical, 0.04-0.06 by 0.03-0.04. Esophagus short, 0.02-0.05 long, bifurcating anteriorly to ventral sucker; intestinal ceca ending at about middle level of hindbody. Ventral sucker median, round, small, slightly posterior to junction of anterior and middle thirds of body, 0.05-0.06 by 0.06-0.07; aperture located on anteroventral margin of ventral sucker, with thick-walled periphery; sucker width ratio 1:0.40–0.50. Ventrogenital sac present, median, small, shallow, enclosing anteroventral half of ventral sucker (Figs. 31 and 32). Ventral invagination present (Figs. 31-33), median, in front of ventral sucker, like tubular pit, 29–32 μm deep by 11  $\mu$ m thick in sagittal sections, usually in-



Figs. 31–34. *Pseudexorchis major*. 31, adult specimen, entire body, ventral view; 32, ventral invagination, ventrogenital sac, ventral sucker, seminal vesicle, and terminal genitalia, ventral view; 33, everted ventral invagination; 34, ovarian complex, dorsal view. cvd, common vitelline duct; ed, ejaculatory duct; gp, genital pore; hd, hermaphroditic duct; lc, Laurer's canal; m, metraterm; mg, Mehlis' gland; o, ovary; od, oviduct; ot, ootype; pp, pars prostatica; sd, sperm duct; sr, seminal receptacle; sv, seminal vesicle; u, uterus; vd, vitelline duct; vgs, ventrogenital sac; vi, ventral invagination; vs, ventral sucker. Scale bars: 0.3 mm in Fig. 31; 0.1 mm in Figs. 32–34.

vaginated but rarely everted, possessing 2 posterior protuberances each with group of small gland cells, sometimes holding one egg in cavity, often with transverse slitlike aperture; its wall thick, anucleate, eosinophilic. Testes double, elliptical to globular, almost symmetrical, submedian, separated, behind intestinal ceca, 0.11-0.16 by 0.06–0.12. Sperm ducts passing transversely, connecting with posterior end of seminal vesicle without common sperm duct (Fig. 32). Seminal vesicle (Figs. 31 and 32) voluminous, bipartite, curved, usually sinistro- but sometimes dextrosubmedian, free in parenchyma, 0.12-0.14 by 0.06-0.08. Cirrus pouch absent. Pars prostatica (Fig. 32) anterointernal to seminal vesicle, free in parenchyma; prostatic cells weakly developed; ejaculatory duct short, uniting with metraterm usually ventrally to anterior chamber of seminal vesicle to form fairly long hermaphroditic duct opening into ventrogenital sac. Genital pore median, on anterior wall of ventrogenital sac. Ovary 3-lobed, submedian, on opposite side of seminal vesicle, behind ventral sucker, 0.09-0.15 by 0.09-0.14; sometimes each lobe indented irregularly. Ovarian complex posterolateral to ventral sucker, on ovarian side of body (Fig. 33). Oviduct short. Seminal receptacle elliptical to globular, posterointernal to ovary, 0.05–0.11 by 0.05-0.08. Laurer's canal almost median, short, running backward. Ootype preovarian, posterolateral to ventral sucker. Mehlis' gland weakly developed, surrounding proximal part of ootype. Uterus passing transversely from ootype to lateral field on ovarian side of body, descending and then ascending there, then crossing body posteriorly to ventral sucker to the other lateral field, similarly descending and then ascending there, usually extending into post-testicular region of body in either lateral field in large, fully mature adults. Metraterm in front of vitelline follicles, running transversely on opposite side of ovary (Figs. 31 and 32). Eggs numerous, lanceolate-oblong, slightly asymmetrical, with small operculum, 35–40 by 17–19  $\mu$ m, embryonated when laid; surface markings seen on eggshell. Vitelline follicles large, 7 each making compact cluster at

level of and dorsally to ovary on either side of body. Excretory vesicle Y-shaped, with arms ending anteriorly between ovary and ventral sucker; excretory pore posterodorsal.

Discussion. Kobayashi (1915) described a new genus and species, Exorchis oviformis (Cryptogonimidae), which is another intestinal digenean of Silurus asotus (syn. Parasilurus asotus) in Japan. Hasegawa (1935a) described a new species, E. major, from the intestine of S. asotus in Japan. Yamaguti (1938) created a new genus, Pseudexorchis, to accommodate E. major as the type species, separating it from the genus Exorchis by the extent of the intestinal ceca and the position of the genital organs. Since Hasegawa's (1935a) original description, no full redescription of P. major has been published.

Pseudexorchis major has a peculiar organ in front of the ventral sucker (Pearson, 2002, 2007; this paper, Figs. 31-33, vi). According to Pearson (2002, 2007), this organ is inside a large ventrogenital sac and "the tubular, eversible invagination of the wall of the ventrogenital sac". He calls it the "ventrogenital invagination". However, the present study has revealed that the invagination is outside a small ventrogenital sac and that it is not associated with either the terminal genitalia or ventrogenital sac. The invagination does not appear to be muscular. Pearson (2002) defined the term gonotyl as "an unbounded, anucleate, typically muscular outgrowth of the wall of the ventrogenital sac". Therefore, I propose that the invagination is referred to as the ventral invagination. In E. oviformis as redescribed by Shimazu (2005), the ventrogenital sac ("[circular] body fold" after him) encloses the genital pore, ventral sucker, and two or three gonotyls ("protrusible muscular structures (gonotyls?)" after him); and the ventral invagination is absent.

Hasegawa (1935a) originally described that a thick-walled genital atrium is located just in front of the ventral sucker and that both ejaculatory duct and metraterm open into the genital atrium side by side. He must have overlooked the ventrogenital sac, hermaphroditic duct, and genital pore; and have mistaken the ventral invagination

for the genital atrium. In other respects, the present specimens agree well with his original description. Hasegawa (1935b) also described the surface markings on the eggshell. Takahashi (1929a) described the ovarian complex in detail.

Because Yamaguti (1938, 1971) follows Hasegawa's (1935a) erroneous original description, the generic diagnoses of the genus *Pseudexorchis* given by him should be amended in part, as follows: ventrogenital sac small, shallow, median, enclosing anteroventral half of ventral sucker, with no gonotyl; hermaphroditic duct fairly long, opening into ventrogenital sac through median genital pore on anterior wall of ventrogenital sac; ventral invagination present, thick-walled, tubular, eversible, not muscular, in front of ventrogenital sac; and uterus descending and then ascending in either lateral field of body, sometimes extending into post-testicular region of body.

Hasegawa (1935a), Takahashi (1929b), and Ito (1956) fully accounted the cercaria and metacercaria of *P. major* from Japan. With regard to the terminal genitalia in the metacercarial stage, they follow the already-mentioned Hasegawa's (1935a) original description of the adult stage. The cercaria of *P. major* was found in the snail *Semisulcospira libertina* from Lake Suwa (Yamaguti, 1938) and *Biwamelania decipiens* (Westerlund) (Pleuroceridae) from Lake Suwa and *Se. libertina* from the Hiroi River (my unpublished data).

This is the first record of the adult of this species from Nagano Prefecture.

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